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## **Crop yield, weed cover and ecosystem multifunctionality are not affected by the duration of organic management**

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Journal Article



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**Crop yield, weed cover and ecosystem multifunctionality are not affected by the duration of organic management**

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26    **Highlights**

- 27    •    Conversion to organic management rapidly shifts agro-ecosystem functioning.
- 28    •    Crop yield, weed cover and biodiversity are unaffected by management duration.
- 29    •    Management practices do not affect ecosystem multifunctionality.

30

31    **Abstract**

32    Organic farming is gaining importance in view of its beneficial effects on soil quality,  
33    environmental performance and biodiversity. However, it is still unclear how organic  
34    management performs over time and whether the duration of organic management  
35    influences crop yield and ecosystem functioning. Here we compared 34 fields in Swiss  
36    farms assigned to four groups: 1) conventionally managed farms; 2) farms in transition  
37    to organic farming (in the 1st–3rd year); 3) farms converted moderately long ago (9–13  
38    years); and 4) farms subjected to long-term organic farming (15–32 years). We selected  
39    one field per farm and examined in two subsequent years whether management  
40    practices (conventional vs. organic farming) and the duration of organic management  
41    affected crop yield, weed cover, soil fertility and biodiversity as well as the overall  
42    system performance, assessed as ecosystem multifunctionality. Maize yield (-6.0%) and  
43    wheat yield (-22.2%) decreased in organic compared to conventional fields. However,  
44    the duration of organic management did not affect crop yield. There was also no effect  
45    of the duration of organic management on weed cover but it was much higher under  
46    organic management, with mean values of 33.0% in organic compared to 2.0% in  
47    conventional fields in maize, and 13.4% compared to 1.2% in wheat, respectively. Soil  
48    fertility and microbial activities were not significantly different between management  
49    practices; only root colonization of arbuscular mycorrhizal fungi increased (+19.7%)  
50    under organic management in wheat. Overall, this study demonstrates a rapid shift of  
51    agro-ecological functions after conversion to organic farming and that the duration of  
52    organic management has no impact on crop yield, weed cover, soil fertility, and  
53    microbial activity.

54  
55    **Keywords:** Organic farming; arbuscular mycorrhizal fungi; management duration;  
56    multifunctionality

## 57    **1. Introduction**

58    In agroecosystems, research is required to develop management strategies that provide  
59    adequate yields while reducing negative environmental impacts in the long term  
60    (Godfray et al., 2010; Godfray and Garnett, 2014; Tilman et al., 2002). Management  
61    practices such as the intensive use of synthetic pesticides and fertilizers in conventional  
62    farming systems can have severe environmental consequences (Pimentel, 2005;  
63    Pimentel et al., 2005) reducing biodiversity, contributing to the production of  
64    greenhouse gases, and causing eutrophication of surface water and drinking water.

65    Changes in land use through agricultural intensification can not only affect biodiversity-  
66    driven ecosystem processes on the local but also on the global scale (Foley et al. 2005).

67        Organic farming has been promoted as a management strategy that could minimize  
68    agriculture's footprint on the environment (Reganold and Wachter, 2016). Even though  
69    the meaning of sustainable agriculture has often been debated in the past, there is little  
70    doubt that sustainable agriculture and many organic farming principles are closely  
71    linked (Rigby and Cáceres, 2001). Organic farming is characterized by no synthetic  
72    pesticide and no synthetic fertilizer use, with beneficial effects on the environmental  
73    performance and social well-being (Reganold and Wachter, 2016). It has been reported  
74    that organic farming has positive effects on soil fertility and biodiversity (Bengtsson et  
75    al., 2005; Mäder et al., 2002; Tuck et al., 2014; Tuomisto et al., 2012), it reduces soil  
76    erosion (Reganold et al., 1987; Seitz et al., 2019) and environmental impacts (Prechsl et  
77    al., 2017) and organic food has lower amounts of pesticide residues (Baker et al., 2002;  
78    Smith-Spangler et al., 2012). In Europe, strict rules are imposed on the use of pesticides  
79    even if certain substances are allowed under organic regulation in other parts of the  
80    world, e.g. rotenone and paraquat. However, organic pesticides such as copper may also  
81    have harmful environmental effects and should therefore be carefully assessed (Bahlai  
82    et al., 2010; Edwards-Jones and Howells, 2001).

83 Despite its beneficial effects, organic agriculture is being critically discussed because  
84 of reduced yield (de Ponti et al., 2012; Ponisio et al., 2015) and reduced yield stability  
85 (Knapp and van der Heijden 2018). Moreover, it is still poorly understood whether the  
86 duration of organic management affects crop yield and ecosystem functions. It is  
87 possible that yield declines over time under organic management due to the  
88 accumulation of pathogens and weeds or gradual decline in soil fertility after conversion  
89 because of reduced fertilizer input (Büchi et al., 2019). Concerns resulting from these  
90 uncertainties may discourage farmers from conversion to organic production (Ferjani et  
91 al., 2010). On the other hand, ecosystem functions that have been shown to be higher  
92 under organic management, such as soil structure (Shepherd et al., 2010), carbon  
93 sequestration (Gattinger et al., 2012), and interactions with beneficial soil symbionts  
94 such as arbuscular mycorrhizal fungi (AMF; Oehl et al., 2003) might further improve,  
95 the longer a field is under organic management. Thus, it is important to investigate to  
96 what extent the duration of organic management affects plant yield, biodiversity and  
97 environmental performance.

98 There is a growing interest in the assessment of ecosystem multifunctionality, which  
99 is the ability of ecosystems to provide multiple functions or services simultaneously  
100 (Manning et al., 2018). One pillar of this approach is the biodiversity-ecosystem  
101 functioning research (Byrnes et al., 2014; Gamfeldt et al., 2008; Hector and Bagchi,  
102 2007; Wagg et al., 2014), the other being the land management research (Bateman et al.,  
103 2013; Manning et al., 2015; Nelson et al., 2009). To our knowledge, the approach has  
104 been mainly used in ecological studies, such as to evaluate biodiversity effects in  
105 grasslands or drylands (Delgado-Baquerizo et al., 2017; Maestre et al., 2012; Soliveres  
106 et al., 2016). In our study, we aimed at applying the concept in investigating to what  
107 extent ecosystem multifunctionality differs between organic and conventional  
108 management and whether it changes over time as a result of continuous organic

109 farming. Further, we wanted to avoid an oversimplified estimation of multifunctionality  
110 and present several multifunctionality scenarios using a modified approach of Allan et  
111 al. (2015) to outline realistic land-use interests (e.g. production, sustainable soils and  
112 cultural services). Our approach of multifunctionality assessment entailed indicator  
113 variables of five ecosystem function categories: productivity (crop yield and weed  
114 cover), soil fertility ( $C_{org}$ , P, K and Mg) and microbial activity (AMF colonization,  
115 microbial biomass C, soil respiration), biodiversity conservation (diversity of weeds,  
116 spiders and root associated fungi) and potential biocontrol of pests (represented by  
117 spider abundance). We varied the weights of the categories to represent  
118 multifunctionality scenarios with different land-use perspectives.

119 To this goal, we established a network in Switzerland comprising a total of 34 fields  
120 in organic and conventional farms. We addressed the following research questions: 1)  
121 Do productivity, soil fertility, soil microbial activity, biodiversity conservation,  
122 potential biocontrol of pests (assessed as spider abundance) and overall ecosystem  
123 multifunctionality differ between conventional and organic fields? 2) And are these  
124 parameters affected by the duration of organic management?

125 We hypothesized that there is 1) A rapid change in plant yield and ecosystem  
126 functioning after the conversion to organic management; 2) Reduced yield and  
127 increased weed cover in organically managed fields; and 3) A continuous enhancement  
128 of biodiversity and soil quality with the duration of organic management.

129

## 130 **2. Materials and methods**

### 131 *2.1. Study sites and study design*

132 We investigated 34 fields included in a farm network in the Cantons of Aargau, Zurich  
133 and Thurgau in Switzerland (Fig. 1). This area is characterized by a damp and mild  
134 climate with Cambisol and Luvisol as the predominant soil types. The farms were

135 divided into four management groups: 1) conventionally managed farms (CONV; 9  
136 farms); 2) organic farms that had been converted recently from conventional  
137 management and therefore still are in transition (TRAN; 9 farms; 1–3 years); 3) organic  
138 farms that were converted moderately long ago (BIO(9); 7 farms; 9–13 years); and 4)  
139 organic farms that had been subjected to long-term organic farming (BIO(15); 9 farms;  
140 15–32 years after conversion to organic farming). Conventional farms were managed  
141 according to the “Proof of Ecological Performance” guidelines of the Swiss Federal  
142 Office for Agriculture (‘Verordnung über die Direktzahlungen an die Landwirtschaft’,  
143 2013), which are the minimum requirements in order to obtain direct payments from the  
144 government and fulfilled by more than 90% of the Swiss farms. Organic farms were  
145 managed according to the guidelines of Bio-Suisse, the Swiss organization for organic  
146 farming (‘<https://www.bio-suisse.ch/>’) and were not allowed to use synthetic fertilizers  
147 as well as pesticides with substances that are synthetically produced and do not occur in  
148 nature.

149         Thirty-four maize (*Zea mays*) fields (one per farm) for ensilage of the whole  
150 plant followed by 24 winter wheat (*Triticum aestivum*) fields on the same parcels were  
151 investigated in 2011 and 2012, respectively (10 parcels were grown with other crops  
152 than wheat in 2012 and were therefore excluded from the 2012 analysis). The fields  
153 were part of a similar crop rotation across farm types with different kind of fodder  
154 production as pre-crops in 2010 (Supplementary Table A4). A total of 13 and 14  
155 different varieties were used for maize and wheat respectively, and this varied  
156 irrespective of management strategy. Fields were ploughed prior to the sowing of maize  
157 and wheat.

158         All measurements except the spider sampling (see below) were taken within a  
159 sampling area, which was a 10 m defined radius around a GPS tagged point  
160 (Supplementary Fig. A.6). A buffer zone of at least 20 m was kept from the border of



161 the field to avoid edge effects, and steep or uneven ground was avoided to gather  
162 representative data. A summary of the variables assessed in each sampling year (e.g. for  
163 maize and wheat) is presented in Table 1 and 2. Questionnaires were collected to assess  
164 management information, such as animal stock, crop rotation, fertilizer inputs and weed  
165 control operations. For the calculation of nutrient inputs applied with organic fertilizers,  
166 we used reference values from GRUDAF (Flisch et al., 2009), the Swiss fertilization  
167 guideline for arable and forage crop production.

168

## 169 *2.2. Yield and weed assessment*

170 For maize, samples for determining yield were taken shortly before harvest between  
171 September and mid-October 2011. In the sampling area, two meters from four rows of  
172 maize were cut and shredded. The dry matter content of maize was determined after  
173 drying a composite sample of about 2 kg at 100 °C for 24 hours. For one of the BIO(15)  
174 maize field, no yield data was available due to heavy hail damage. For wheat, yield  
175 samples were collected in July 2012 from four randomly chosen sub-plots (0.6 x 0.4 m)  
176 within the sampling area. A subsample of grains was oven-dried at 100 °C for 24 hours  
177 to calculate the dry matter (DM) grain yield in decitonne (dt) per hectare. Weed species  
178 and the relative ground cover in percentage of each species were determined visually at  
179 three randomly selected areas within the sampling area; 0.75 x 0.75 m for maize in July  
180 2011 and 1 x 1 m for wheat in June 2012. One wheat field (TRAN) had to be excluded  
181 from the weed assessment as there was only lodged wheat in the sampling area. For the  
182 analysis, we averaged data of the three sub-plots.

183

## 184 *2.3. Soil chemical properties*

185 In early March 2012, a composite soil sample was collected in each sampling area at 20  
186 cm depth and stored at 4° C until further processing. Soils were analyzed according to

the reference methods of the Swiss Federal Institutes of Agricultural Research (Eidgenössische Forschungsanstalten FAL, RAC, 1996). For texture, clay and silt contents were assessed in water suspension aliquots by sedimentation analysis. Soil pH was measured from an aqueous soil suspension and a water to soil ratio of 1:2.5 based on the potentiometric measurement of hydrogen ion activity. The organically bound carbon ( $C_{org}$ ) was oxidized in excess by the addition of potassium dichromate ( $K_2Cr_2O_7$ ). The remaining  $K_2Cr_2O_7$  solution was titrated back with a Fe(II) solution. Given the redox reaction,  $C_{org}$  (mass-percentage) can be determined, which is proportional to the consumed  $K_2Cr_2O_7$ .

Nutrient contents were determined from soil samples collected in July 2012 to allow two months of time after the last fertilizer application. Soils were 2 mm sieved prior to analysis. To extract easily soluble phosphorus (P) and potassium (K) we used a ratio of soil to  $CO_2$ -saturated water of 1:2.5. The extracted P was converted to phosphor molybdenum blue with ammonium molybdate in acidic solution. The resulting blue coloration was determined photometrically at a wavelength of 750 nm. P was determined by flame emission at a wavelength of 769.9 nm. The easily exchangeable magnesium (Mg) was extracted from the soil with a  $CaCl_2$  solution. This method is based on the ion exchange of Mg with calcium (Ca). The ratio of soil to 0.0125 M  $CaCl_2$  solution was 1:10. Mg was determined by atomic absorption spectroscopy at a wavelength of 202.6 nm.

#### 2.4. Soil biological properties

To assess basal respiration, soils were pre-incubated for seven days at 50% water holding capacity to stabilize microbial communities after sample preparation. Before the actual measurement, soils were incubated in a closed system with a NaOH solution for 24 hours and transferred to a new bottle to absorb the emitted  $CO_2$  in a NaOH

213 solution over 72 hours. The resulting  $\text{Na}_2\text{CO}_3$  was precipitated with  $\text{BaCl}_2$  and the  
214 unused NaOH was determined by titration with HCl (Isermeyer, 1952; Jäggi, 1976).  
215 Soil microbial biomass carbon (MBC) was measured by chloroform-fumigation-  
216 extraction according to Vance et al., (1987). Fresh soil samples corresponding to 20 g  
217 dry soil, were fumigated with chloroform for 24 hours. Organic C content was  
218 measured by infrared spectrometry after combustion at 850 °C (DIMATOC® 2000,  
219 Dimatec, Essen, Germany).

220

#### 221 *2.5. AMF colonization and the overall root associated fungal communities*

222 In August 2011, a pooled sample of fine roots from five maize plants, cut in pieces of  
223 about 2 cm length, was stored in 50% EtOH for the analysis of root colonization by  
224 AMF. Likewise in June 2012, roots were collected from six wheat plants and an  
225 additional subsample was preserved in liquid nitrogen and stored at – 80 °C for  
226 subsequent molecular analyses of fungal communities. The percentage of root  
227 colonization by AMF was assessed following the procedure of Vierheilig *et al.* (1998).  
228 In brief, roots were cleared in 10% KOH in a water bath at 80 C° for 25 min and stained  
229 with a 5% ink-vinegar solution for 15 min. Roots were prepared on a microscopy slide  
230 and colonization was measured with a light microscope at a magnification of x 200  
231 using a modified line-intersection method for a hundred intersections per sample.  
232 (McGonigle et al., 1990).

233 A detailed description of sample preparation and bioinformatic analysis of root  
234 associated fungal communities in wheat can be found in the study of Verbruggen et al.  
235 (2014). In brief, DNA was extracted from 7 to 10 mg lyophilized roots using the  
236 Dneasy Plant Mini Kit. PCR was conducted using the Firepol DNA polymerase (Solis  
237 Biodyne, Tartu, Estonia) and the general fungal primers ITS1F (Gardes and Bruns,  
238 1993) and ITS4 (White et al., 1990). Amplicons were sequenced on four 1/8 plate-

239 regions using the Roche 454 FLX Titanium pyrosequencing (Roche, Branford, CT,  
240 USA). We obtained 519,110 reads with an average read length of 413.79 bp. Reads  
241 were filtered, reads shorter than 200 bp were excluded followed by clustering and  
242 classification of operational taxonomic units (OTUs). All samples were resampled to  
243 the minimum read number to account for differences in sequencing depth, leaving a  
244 total of 394 fungal OTUs.

245

## 246 2.6. *Spiders*

247 Spider data were collected between July and August 2011 in 31 out of 34 maize fields  
248 using a suction sampling method (Jeanneret et al., 2012) with a modified vacuum  
249 shredder powered by a two-stroke engine (Stihl 86-D). Five sub-samples per field were  
250 randomly collected (Supplementary Fig.A6), pooled and stored in 70% EtOH. Details  
251 on the sampling procedure are given in Supplementary data A.1. Spiders were attributed  
252 the family and if possible the genus using Roberts (1995) and then identified as  
253 morphospecies which are reliable surrogates for taxonomic unit species for studying  
254 patterns of species richness and diversity (Oliver and Beattie, 1996). In our analysis, we  
255 averaged the three sampling rounds to get one mean value for spider abundance per  
256 species.

257

## 258 2.7. *Multifunctionality assessment*

259 The indicator variables used to assess multifunctionality (16 in total, see Table 3) were  
260 classified into five ecosystem function (EF) categories, namely: 1) “productivity”, 2)  
261 “soil fertility”, 3) “soil microbial activity”, 4) “biodiversity conservation”, and 5)  
262 “potential biological control of pests”. A detailed description of the classification and  
263 calculation of z-scores can be found in Supplementary data A.2. We calculated  
264 standardized z-scores for the 16 indicator variables (Wagg et al., 2014) and averaged

265 them according to Table 3 to build the five EF categories. For the purpose of expressing  
 266 different management priorities (e.g. yield or soil protection oriented), Allen et al.  
 267 (2015) used specific sets of functions and services with different weightings in order to  
 268 represent different land-use perspectives: 1) “production only”; 2) “sustainable soils”;  
 269 3) “sustainable soils and crops”; 4) “equal weight multifunctionality”; and 5) “cultural  
 270 multifunctionality”. Similar to Allan et al. (2015) but adapted to our context, we  
 271 calculated four multifunctionality indices by averaging z-scores of the five EF  
 272 categories after giving them different weightings: (M1) “production” with 50% of the  
 273 weight for “productivity” and 50% weight for the remaining variables; (M2)  
 274 “sustainable soils” with 25% of the weight for each “productivity “, “soil microbial  
 275 activity” and “soil fertility” and 25% weight for the remaining variables; (M3) “equal  
 276 weight multifunctionality” weighs all EF categories equally; and (M4) “biodiversity”  
 277 with 50% of the weight on the EF category “biodiversity” (Fig. 2). The scenarios  
 278 indicate a gradient from prioritizing agricultural production (M1) to biodiversity (M4)  
 279 via a neutral scenario (M3).

280

## 281 2.8. Statistical analyses

282 In total, 14 and 20 response variables were individually analyzed for maize and wheat  
 283 fields, respectively (Tables 1, 2). All statistical analyses were performed using R  
 284 version 3.5.0 (R Core Team, 2018). As the assumptions for parametric statistics were  
 285 violated for nearly all variables, we tested the effects of the management groups i.e.  
 286 CONV, TRAN, BIO(9) and BIO(15), on the examined variables for significant  
 287 differences ( $\alpha = 0.05$ ) using the non-parametric Kruskal-Wallis test. Multiple  
 288 comparisons of group means were performed using the *kruskalmc* function from the  
 289 *pgirmess* package. Weed cover was averaged over the three quadrats and the Shannon-  
 290 Wiener index of diversity was calculated using the *diversity* function available in the

291 *vegan* package. For the analysis of fungal communities, read numbers of operational  
292 taxonomic units (OTUs) were rarefied for each sample to the minimum read number  
293 obtained in all samples using the *phyloseq* package. Alpha diversity of the rarefied data  
294 was calculated for each field using the *estimate\_richness* functions. We performed a  
295 principal component analysis (PCA) using the *prcomp* function to visualize the  
296 variables included in the multifunctionality analyses (Table 3) and the management  
297 groups in the multidimensional space. To test whether variances in our data can be  
298 explained by the management groups, we run a permutational multivariate ANOVA  
299 (PERMANOVA) with the Euclidean distance matrices using the *adonis* function with  
300 999 permutations. Pairwise correlations were tested for both study years using the *cor*  
301 function based on spearman rank correlation coefficient and visualized in a correlogram  
302 using the *corrplot* function. Management group effects on multifunctionality indices  
303 were analyzed the same way as the individual variables and visualized with boxplots.  
304 Simple linear regressions were further used to establish the relation of yield and  
305 multifunctionality with organic management duration. Relationships were visualized by  
306 scatterplots including the regression lines. If not stated otherwise, graphs were created  
307 using the *ggplot2* package.

308

### 309 **3. Results**

#### 310 *3.1. Crop yield and weed cover*

311 Yields of organic fields were on average 6.0% lower than in conventional fields for  
312 maize (Chi square = 1.191, df = 1, p = 0.276) and 22.2% for wheat respectively (Chi  
313 square = 6.947, df = 1, p = 0.008). Wheat and maize yield was not affected by the  
314 duration of organic management (Fig. 3). Fields in transition to organic agriculture (1-3  
315 years) had similar yields as fields managed organically between 9 and 32 years for  
316 maize (Chi square = 0.556, df = 1, p = 0.456) and wheat (Chi square = 0.042, df = 1, p =

0.838). Moreover, weed cover showed similar patterns for both crops with clearly higher weed cover in organic fields, including those under conversion, and similar to yield, weed cover was not affected by the duration of organic farming (Figure 3).

### 3.2. *Effect of organic farming on biodiversity, soil microbial activity and soil fertility*

Weed species richness varied depending on the treatment and was positively affected by organic management in both crops (Tables 1, 2). Weed diversity, reflected by the Shannon index, was on average significantly increased in organic fields in maize (+215.3%; Chi square = 14.53, df = 1, p = < 0.001) and wheat (+92.9%; Chi square = 5.083, df = 1, p = 0.024). Species richness, evenness and diversity of both spiders and root associated fungi were neither affected by the management nor by the duration of organic farming (Tables 1, 2). Spider abundance was positively correlated with weed species richness (Supplementary Fig. A.7).

AMF colonization in maize was similar in conventional compared to organically managed plots (+1.6% in organic; Chi square = 0.152, df = 1, p = 0.696). AMF colonization in wheat was significantly higher in BIO(9) than in conventional fields, indicating an overall positive effect of organic farming practices on AMF (+19.7%; Chi square = 5.083, df = 1, p = 0.024; Table 2). Both MBC and soil respiration did not differ significantly between management groups, and neither did C<sub>org</sub>, although it tended to be higher in organically managed fields (+15.4% in organic; Chi square = 2.624 df = 1, p = 0.105). P and Mg in the soil did not respond to management while K was clearly higher in BIO(15) soil compared to TRAN, BIO(9) but not conventionally managed fields (Table 2).

### 3.3. *Effect of organic farming on ecosystem multifunctionality*

A PCA revealed a clustering of conventional farms (Fig. 4). PERMANOVA further showed a statistically significant difference between conventional and organic fields, including fields in transition for maize ( $F_{1,20} = 2.281$ ,  $p = 0.005$ ) and wheat ( $F_{1,20} = 2.223$ ,  $p = 0.006$ ). The indicator variables best explaining differences among the treatments in the maize data were spider abundance, weed species richness and spider evenness (principal component PC1) as well as yield, weed cover and spider richness (principal component PC2). For wheat, it was MBC, soil respiration and  $C_{org}$  for PC1 and yield, weed species richness and weed cover for PC2.

Our analysis did not reveal significant differences between management groups on the four multifunctionality indices, namely “(M1) production”, “(M2) sustainable soils”, “(M3) equal weight multifunctionality” and “(M4) biodiversity” (Fig. 5; Supplementary A.8), despite that the EF category “productivity” was higher in conventional fields. Although differences were not significant, mean multifunctionality of the organic farms in M1 was the highest in the long-term group BIO(15) and on a similar level as the conventional farm group. Moreover, for M2-M4, multifunctionality tended to increase with organic management duration and the long-term organic farms showed the highest mean values. Our results demonstrated a response of crop yield, weed cover and environmental performance to the conversion from organic to conventional farming but no change along with organic farming duration.

361

## 362 **4. Discussion**

### 363 *4.1. Yield and weed cover are determined by management practice but not management* 364 *duration*

365 In agricultural research, yield is one of the key factors to consider when evaluating  
366 different food production systems. In our study, we were able to explore two important  
367 questions related to crop yield under organic farming: 1) Do yield and weed cover differ



368 between conventional and organic fields? and 2) Are the two parameters affected by the  
369 duration of organic management? On average, we found 6.0% lower yield for maize and  
370 22.2% for wheat, respectively (Tables 1, 2) which is in line with earlier reports from  
371 Switzerland that showed 10% and 20% lower yield for maize and wheat, respectively  
372 (Jossi et al., 2009; Zihlmann et al., 2010). While a range of studies investigated organic  
373 yield gaps, very few tested whether the duration of organic management altered plant  
374 yield and they largely focused on the transitional period (e.g. Gopinath et al., 2008;  
375 Martini et al., 2004). Our study shows no significant decline of maize- and wheat yields  
376 with increasing duration of organic management. Thus, our results indicate, that under  
377 Swiss farming conditions, farmers interested in a conversion to organic farming should  
378 not be concerned with a significant gradual decline of yield. However, in certain cases,  
379 the yield gap can be more pronounced, for example Larsen et al. (2014) observed a  
380 reduction of more than 50% maize yield in organic compared to conventional  
381 treatments. Moreover, the findings of Schrama et al. (2018) show that closing the yield  
382 gap between organic and conventional farming can be a question of time and that  
383 organic farms potentially result in higher spatial stability due to slow changes in soil  
384 properties.

385        Besides yield losses, weed cover is a major problem under organic management.  
386 Unlike conventional systems that allow the use of herbicides as an effective weed  
387 control strategy, organic farmers have to pursue alternative weed control strategies such  
388 as manual removal, thermal methods or tillage. Although soil cover by weeds generally  
389 increased after conversion to organic farming, the weed abundance did not significantly  
390 increase with time after conversion from conventional to organic management. In the  
391 long-term organic farm group BIO(15), values of weed cover were between 5.3% and  
392 78% for maize and between 4.2% and 25.1% for wheat, respectively. This large range  
393 can partly be explained by the fact that weed cover per se is strongly influenced by site

394 conditions and other context dependent features such as weed control strategies and  
395 weed tolerance of farmers.

396 Weed cover was negatively correlated with wheat but not maize yield, which  
397 could explain why the yield gap was much stronger for wheat (-22,2%) than maize (-  
398 6%). This observation could indicate a stronger crop-weed competition for wheat  
399 compared to maize, and we encourage future research to test this assumption and  
400 investigate how crops differ in their competing ability with weeds. Another explanation  
401 that needs to be tested, is that a large amount of farm manure (mostly slurry and also in  
402 conventional farms) is generally applied to maize and that mineralization time fits better  
403 with crop vegetation, thus reducing fertilization gap between organically and  
404 conventionally managed fields.

405

#### 406 *4.2. Organic farming maintains soil fertility*

407 Preservation of fertile soils is essential for sustainable food production system but it is  
408 still debatable whether soil nutrient contents can be kept at an optimum level over a  
409 long period of time under organic farming. Previous research showed reduced levels of  
410 P and K in soils under organic management, which was mainly explained by reduced  
411 fertilizer inputs as well as foregoing the use of mineral fertilizers (Gosling and  
412 Shepherd, 2005). In our comparison of plant available soil nutrients of organic and  
413 conventional farms there was no difference in P and Mg contents between both farming  
414 systems. Surprisingly for K, the long-term organic farm group BIO(15) showed the  
415 highest levels in our study. This can be explained by the large amount of nutrient-rich  
416 organic fertilizers, mainly in the form of potassium oxide (K<sub>2</sub>O) rich cattle manure  
417 applied on organic farms. Thus, nutrient supply should not discourage farmers from a  
418 conversion to organic management, especially for mixed farms. This study did not  
419 assess the level of mineral N (N<sub>min</sub>) in the soil, which should be considered in future

work. However, we evaluated fertilizer inputs and the results show significantly lower N-fertilization in BIO(15) than CONV fields for maize, and in BIO(9) and BIO(15) compared to CONV fields for wheat (Tables 1, 2; Supplementary data A.3). Moreover,  $C_{org}$ , an important indicator for soil quality (Liu et al., 2006; Reeves, 1997), was also unaffected by management (e.g. slight higher values (+15.4%) under organic management). Higher  $C_{org}$  values in organic compared to conventional farming systems have been reported in literature (Reganold, 1995, 1988), an earlier Swiss study also found no significant differences in  $C_{org}$  between organic and conventional fields (Oberholzer et al., 2009). It is important to note that most Swiss farms are mixed farms, with dairy and arable cropping production, that can use their cattle manure on arable lands (Fliessbach et al., 2007). As a result, not only organically but also conventionally managed fields are supplied with organic fertilizer, which may be a key factor resulting in less pronounced disparities in  $C_{org}$  in Swiss agroecosystems.

#### *4.3. Soil microbiology only differs in AMF root colonization*

Soil microbial biomass is sensitive to agricultural management practices (Wardle, 1992) and can be used as an indicator for soil health and environmental sustainability (Singh and Gupta, 2018). We did not find a significant difference in MBC between the management groups in our study, although organically managed fields tended to have higher MBC (+23.2%). More diverse crop rotations and the widespread application of farmyard manure of conventional farms in comparison to other countries may partly explain the difference between our results and that found in field experiments or other countries (Araújo et al., 2008; Gunapala and Scow, 1998; Mäder et al., 2002; Tu et al., 2006). Through a wider range of field sites, our results support findings of Swiss study in which there was no difference in microbial biomass between the two farming systems in a field experiment (Fliessbach et al., 2007).

Arbuscular mycorrhizal fungi (AMF) are important bioindicators in agricultural systems because they influence plant growth and are sensitive to changes in land use type and intensity (Oehl et al., 2011; Verbruggen et al., 2010). While there was no management effect on maize, we found a positive impact of organic farming on AMF root colonization in wheat (Tables 1, 2). In contrast to a comparable study performed in the Netherlands (Verbruggen et al., 2010), we could not find a response of AMF colonization to the duration of organic farming. However, overall differences between organic and conventional management are stronger in the Netherlands than in Switzerland, e.g. many maize fields of the Dutch study were under long-term monocropping. Moreover, the strict Swiss management regulations, e.g. crop rotation and reduced fertilizer use, also in conventionally managed fields, may explain why soil respiration, MBC and AMF root colonization did not greatly differ between conventional and organic farming.

459

#### 4.4. No increase of biodiversity along with management duration

In our study, we only found positive effects of organic farming on weed species richness and diversity but not on overall spider- or root associated fungal communities and they were not affected by management duration. Spiders, which are important for biological pest control in agricultural fields (Marc and Canard, 1997; Samu and Szinetár, 2002; Sunderland and Samu, 2000) did not differ in their abundance or diversity between farming practices. Though Hole et al. (2005) concluded that most of the reviewed studies reported higher spider abundances under organic management, differences were not always statistically significant across sites and years. In accordance with earlier studies showing that the number and variety of spiders mainly depend on the habitat structure (Hole et al., 2005; Jeanneret et al., 2003; Samu and Szinetár, 2002), we found that spider abundances were positively correlated with weed species richness.

472 Even though we expected more diverse spider communities in organically managed  
473 fields, e.g. through higher crop diversification, spiders also depend to a large extent on  
474 the surrounding landscape (Sunderland and Samu, 2000).

475         Root associated fungal communities did not differ between management groups.  
476 Our finding is in accordance with Hole et al. (2005) who observed a little difference in  
477 fungal communities between organic and conventional systems. Banerjee et al. (2019)  
478 also found no differences in fungal diversity between organic and conventional fields.  
479 However, in that study organically managed fields harbored a much more complex  
480 fungal network with more keystone taxa. Furthermore, Verbruggen et al. (2014)  
481 discovered that *Sebacinales* were only present in organic fields of our farmer network.  
482 This finding is particularly interesting because these endophytic fungi are known to  
483 form beneficial interactions with their host plants, involving enhanced resistance to  
484 abiotic and pathogen stress (Michael et al., 2016). Future research should therefore test  
485 the potential of *Sebacinales* to serve as bio-indicators for sustainable land use  
486 (Verbruggen et al., 2014) and complement other already well-known indicator groups  
487 such as AMF (Jansa et al., 2014; Oehl et al., 2011). Comprehensive meta-analyses from  
488 Bengtsson et al. (2005) and Tuck et al. (2014) reported an overall positive effect of  
489 organic farming on species richness and abundance but stated that the effects mostly  
490 depended on organism identities and landscape types, and that differences are most  
491 pronounced in intensively managed systems. Across a large range of European farming  
492 systems and environmental conditions, Schneider et al. (2014) also showed an overall  
493 higher species richness (plants, earthworms, spiders, bees) in organic systems than in  
494 conventional ones at plot scale but not farm scale. Our study contributes to the  
495 conclusion that effects of organic farming on biodiversity depend on the organisms  
496 under study as well as on the scale investigated.

497

#### 498    *4.5. Agro-ecological functioning and multifunctionality across management groups*

500    Principal component analysis (Fig. 4) showed that organically managed fields  
501    including the fields recently converted to organic management clustered together while  
502    conventional fields formed a separate cluster. This outcome suggests a rapid shift of  
503    agro-ecological functions after a conversion to organic farming as fields of farms in  
504    transition are more similar to long term organic than to conventional managed fields.  
505    The clustering of conventional fields versus organic fields was mainly determined by  
506    yield, weed abundance and weed species richness. Yield and weeds can respond  
507    quickly to a change of management (e.g. because under organic management weeds are  
508    no longer suppressed with herbicides and yield is reduced because the amount of  
509    directly available nutrients is lower while mineral fertilizers cannot be applied directly  
510    when crop requirements for fertilizers are highest). Future studies investigating agro-  
511    ecological functioning of conventional and organically managed fields should also  
512    focus on variables that may respond more slowly to a transition to organic farming (e.g.  
513    soil quality indicators not assessed in this study) and include more fields, if possible  
514    fields that have been under organic management for over 50 years.

515    In our multifunctionality assessment, we did not find clear management effects  
516    on the four ecosystem multifunctionality scenarios. This could be explained by several  
517    factors. First, there was substantial variations between individual fields within  
518    treatments. Second, differences between treatments were apparently not strong enough  
519    and opposing effects of different variables (e.g. higher yield, but lower weed diversity  
520    in conventionally managed fields, and opposite effects in organically managed fields)  
521    cancelled out effects. Moreover, although we analyzed a wide range of indicator  
522    variables (16 in total), it is unfeasible to assess all possible functions (Manning et al.,  
523    2018) and we might have missed important variables. The scenario that focuses on  
524    production showed no further decrease of crop yield along with organic management

524 duration. The remaining scenarios, namely sustainable soils, equal weight and  
525 biodiversity, all showed increasing multifunctionality with ongoing organic  
526 management, which implied a positive trend of the impact of ongoing organic farming  
527 on the overall environmental performance when emphasizing on alternative land use  
528 objectives rather than productivity alone. This increase could be driven by the higher  
529 soil-K contents resulting from the use of organic fertilizer that is rich in K and widely  
530 applied on organic farms, as well as the higher weed species richness and overall AMF  
531 colonization we observed in organically managed fields. To better understand how  
532 organic management influences ecosystem multifunctionality in the long-term, we need  
533 further studies that assess a large number of fields over multiple years and test a  
534 comprehensive set of variables that represent a wide range of ecosystem functions and  
535 services.

536

## 537 **5. Conclusion**

538 A range of studies compared yield, biodiversity and environmental performance of  
539 organic versus conventionally managed fields. However, it is still unclear whether the  
540 duration of organic management affects plant yield and ecosystem functions. Our  
541 investigation demonstrated that crop yield, weed cover, soil fertility, biodiversity and  
542 potential biocontrol of pest through spiders are not affected by the duration of organic  
543 management. Moreover, we found positive effects of organic farming on AMF root  
544 colonization, weed species richness and soil-K contents. The present study may help  
545 reduce concerns of farmers related to long-term organic farming. Finally, our results  
546 may contribute to a better understanding of the role of different farming systems on the  
547 environmental performance.

548

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## Figure captions

**Fig. 1.** Map of Switzerland with the 34 study sites in the Cantons of Aargau, Zurich and Thurgau. One crop field was selected at each study site and was assigned to one of the four management groups: 1) conventionally managed farms (CONV); 2) fields in transition to organic farming (in the 1st–3rd year; TRAN); 3) fields converted moderately long ago (9–13 years; BIO(9), 4) or fields that had been subjected to long-term organic farming (15–32 years, BIO(15). [Source: Swisstopo (2018)]

**Fig. 2.** Comparison of four different multifunctionality scenarios representing different land-use perspectives: (M1) “production” with 50% of the weight for “productivity” and 50% weight for the remaining variables; (M2) “sustainable soils” with 25% of the weight for each “productivity”, “soil microbial activity” and “soil fertility” and 25% weight for the remaining variables; (M3) “equal weight multifunctionality” weighs all EF categories equally; and (M4) “biodiversity” with 50% of the weight on the EF category “biodiversity”.

**Fig. 3.** Linear regression plots showing the relationships between the duration of organic management and maize yield (A), wheat yield (B), weed cover in maize (C) and weed cover in wheat (D). Fields were assigned to four management groups: 1) conventionally managed farms (CONV); 2) fields in transition to organic farming (in the 1st–3rd year; TRAN); 3) fields converted moderately long ago (9–13 years; BIO(9), 4) or fields that had been subjected to long-term organic farming (15–32 years, BIO(15). The duration of organic management is expressed as the number of years since the conversion from conventional to organic management. The black line indicates the linear regression line and the gray region the 95% confidence interval. Conventional farms were plotted for the comparison but are not included in the regression analysis.

**Fig. 4.** Principal component analysis (PCA) comparing maize (A) and wheat (B) fields under conventional management (CONV); fields in transition to organic management (in the 1st–3rd year; TRAN), fields managed organically between nine and 13 years (BIO(9)) or fields that had been subjected to long-term organic farming (15–32 years, BIO(15). The same variables (eight for maize and 13 for wheat) were used to calculate ecosystem multifunctionality. Variables were standardized prior to the application of PCA. The principal component axes 1 and 2 (PC1 and PC2 together explained 57.3% of the total variation in the response of the maize fields and 53.2% of the wheat fields, respectively. The four ellipses represent 68% confidence around each management group. Variable abbreviations are: weed %= weed cover, P= soil P, K=soil K, Mg=soil Mg, AMF %=AMF colonization, resp=respiration, weed S= weed species richness, weed J= weed species evenness, spid S=spider species richness, spid J=spider species evenness, fungi S=OTU richness of root associated fungi, fungi J=OTU richness of root associated fungi, spid abund=spider abundance.

**Fig. 5.** Multifunctionality in response to the different management and the four different scenarios: (A) M1) “production” with 50% of the weight for “productivity” and 50% weight for the remaining categories; (B) M2) “sustainable soils” with 25% of the weight for each “productivity”, “soil microbial activity” and “soil fertility” and 25% for the remaining categories; (C) M3) “equal weight multifunctionality” where all EF categories are weighted equally; and D) M4) “biodiversity” with 50% of the weight on the EF category “biodiversity” and 50% for the remaining categories (Fig. 2). Each field

929 was assigned to one of the four management groups: 1) conventionally managed farms  
930 (CONV); 2) fields in transition to organic farming (in the 1st–3rd year; TRAN); 3)  
931 fields converted moderately long ago (9–13 years; BIO(9), 4) or fields that had been  
932 subjected to long-term organic farming (15–32 years, BIO(15). Bold lines represent  
933 medians, black crosses the means, boxes the first and third quantiles. Ns indicates non-  
934 significant differences among treatments at an alpha value of 0.05.

935 **Table 1.** Mean, standard errors (SE), sample size (N) and the statistical output of the Kruskal-Wallis test for the assessed variables in maize (N=34).  
936 P-values in bold indicate significant effects at  $P = 0.05$ . The farms were divided into four groups: 1) conventionally managed farms (CONV; 9  
937 farms); 2) organic farms that had recently been converted (TRAN; 9 farms; 1–3 years); 3) moderately long ago (BIO(9); 7 farms; 9–13 years), 4) or  
938 had been subjected to long-term organic farming (BIO(15); 9 farms; 15–32 years). Variables included in the multifunctionality assessment are  
939 indicated as indicator variables. Different letters after the mean indicate significant differences among management groups, at an alpha rejection  
940 value set to 0.05.

Variable	Indicator variable	CONV			TRAN			BIO(9)			BIO(15)			941	942
		Mean	SE	N	Mean	SE	N	Mean	SE	N	Mean	SE	N	H(df=3)	p-value
Yield (dt DM/ha)	yes	207.089	5.403	9	196.478	9.816	9	190.229	6.291	7	196.550	12.089	8	2.590	0.459
Weed cover (%)	yes	2.033 <b>a</b>	0.661	9	27.711 <b>b</b>	5.714	9	33.229 <b>b</b>	10.046	7	38.222 <b>b</b>	9.257	9	17.832	< <b>0.001</b>
Weed species richness ( <i>S</i> )	yes	1.889 <b>a</b>	0.484	9	5.667 <b>b</b>	0.833	9	7.286 <b>b</b>	1.107	7	8.889 <b>b</b>	1.047	9	18.982	< <b>0.001</b>
Weed species evenness ( <i>J</i> )	yes	0.518	0.150	7	0.750	0.038	8	0.707	0.034	7	0.702	0.045	9	1.298	0.730
Weed diversity ( <i>H</i> )	no	0.428 <b>a</b>	0.151	9	1.196 <b>b</b>	0.177	9	1.365 <b>b</b>	0.171	7	1.491 <b>b</b>	0.130	9	15.309	<b>0.002</b>
Spider species richness ( <i>S</i> )	yes	6.111	0.992	9	6.875	0.833	8	5.830	1.352	6	6.500	0.627	8	0.662	0.882
Spider species evenness ( <i>J</i> )	yes	0.828	0.036	8	0.745	0.052	8	0.710	0.079	6	0.776	0.042	8	1.688	0.640
Spider diversity ( <i>H</i> )	no	1.373	0.212	9	1.360	0.099	8	1.090	0.156	6	1.415	0.102	8	2.888	0.409
Spider diversity (Chao1)	no	18.850	2.858	8	17.014	3.233	8	20.290	6.763	5	19.774	3.934	8	0.485	0.922
Spider abundance	yes	6.889	1.399	9	11.000	1.783	8	11.000	4.435	6	7.375	2.112	8	3.639	0.303
AMF colonization (%)	yes	43.333	4.503	9	35.556	3.379	9	43.143	6.759	7	46.889	3.549	9	3.824	0.281
Fertilizer-N (kg N <sub>soluble</sub> /ha)	no	152.000 <b>a</b>	16.562	9	115.111 <b>ab</b>	20.259	9	90.800 <b>ab</b>	7.851	5	84.875 <b>b</b>	14.593	8	8.255	<b>0.041</b>
Fertilizer-P (kg P <sub>2</sub> O <sub>5</sub> /ha)	no	102.222	18.125	9	124.222	15.259	9	107.400	31.384	5	102.625	14.931	8	1.792	0.617
Fertilizer-K (kg K <sub>2</sub> O/ha)	no	226.444	38.639	9	286.222	37.034	9	289.400	15.224	5	216.142	25.078	7	4.719	0.194



943 **Table 2.** Mean, standard error (SE) and sample size (N) and the statistical output of the Kruskal-Wallis test for the assessed variables in wheat fields  
944 (N=24). P-values in bold indicate significant effects at  $P = 0.05$ . The farms were divided into four groups: 1) conventionally managed farms  
945 (CONV; 7 farms); 2) organic farms that had recently been converted (TRAN; 6 farms; 1–3 years); 3) moderately long ago (BIO(9); 5 farms; 9–13  
946 years), 4) or had been subjected to long-term organic farming (BIO(15); 6 farms; 15–32 years). Variables included in the multifunctionality  
947 assessment are indicated as indicator variables. Different letters after the mean indicate significant differences among management groups, at an  
948 alpha rejection value set to 0.05.

Variable	Indicator variable	CONV			TRAN			BIO(9)			BIO(15)			H(df=3)	p-value
		Mean	SE	N	Mean	SE	N	Mean	SE	N	Mean	SE	N		
Yield (dt DM/ha)	yes	64.371	4.093	7	50.317	3.473	6	50.460	3.6140	5	49.517	3.500	6	6.962	0.073
Weed cover (%)	yes	1.233 <b>a</b>	0.596	7	11.767 <b>b</b>	2.367	6	15.420 <b>b</b>	11.176	5	13.450 <b>b</b>	3.557	6	12.301	<b>0.006</b>
Weed species richness ( <i>S</i> )	yes	3.143 <b>a</b>	0.595	7	11.000 <b>b</b>	1.238	6	8.400 <b>ab</b>	1.720	5	9.667 <b>b</b>	1.229	6	14.468	<b>0.002</b>
Weed species evenness ( <i>J</i> )	yes	0.680	0.100	6	0.527	0.083	6	0.687	0.096	5	0.491	0.055	6	2.054	0.561
Weed diversity ( <i>H</i> )	no	0.668	0.174	7	1.262	0.210	6	1.263	0.267	5	1.116	0.107	6	5.218	0.157
OTU richness ( <i>S</i> ) of root associated fungi	yes	57.430	6.679	7	57.167	3.554	6	58.600	4.250	5	54.000	5.994	6	0.660	0.883
OTU evenness ( <i>J</i> ) of root associated fungi	yes	0.607	0.043	7	0.531	0.053	6	0.586	0.065	5	0.471	0.072	6	3.059	0.383
OTU diversity ( <i>H</i> ) of root associated fungi	no	2.453	0.220	7	2.146	0.225	6	2.397	0.303	5	1.903	0.331	6	2.223	0.527
Microbial biomass C (mg C/kg soil)	yes	551.587	104.256	7	612.900	78.740	6	646.340	104.600	5	774.567	99.445	6	2.369	0.500
Respiration (mg CO <sub>2</sub> -C/kg soil*h)	yes	0.569	0.085	7	0.6708	0.071	6	0.669	0.094	5	0.797	0.101	6	3.115	0.374
AMF colonization (%)	yes	27.857 <b>a</b>	5.705	7	47.833 <b>ab</b>	2.088	6	54.000 <b>b</b>	2.074	5	42.000 <b>a</b>	3.183	6	13.905	<b>0.003</b>
C <sub>org</sub> (%)	yes	1.817	0.210	7	1.943	0.186	6	2.134	0.132	5	2.218	0.154	6	3.937	0.268
Soil K (mg/100g soil)	yes	2.657 <b>ab</b>	0.324	7	1.283 <b>b</b>	0.221	6	1.640 <b>b</b>	0.434	5	4.867 <b>a</b>	1.147	6	14.192	<b>0.003</b>
Soil P (mg/100g soil)	yes	11.043	1.149	7	6.200	2.019	6	8.9200	1.605	5	13.483	2.794	6	6.569	0.087
Soil Mg (mg/100g soil)	yes	13.943	3.587	7	15.783	2.362	6	21.100	7.417	5	19.867	5.197	6	2.048	0.563
Soil pH March	no	6.857	0.208	7	6.767	0.329	6	6.660	0.304	5	7.200	0.200	6	2.196	0.533
Soil pH July	no	6.700	0.129	7	6.567	0.171	6	6.500	0.084	5	6.817	0.091	6	4.197	0.241

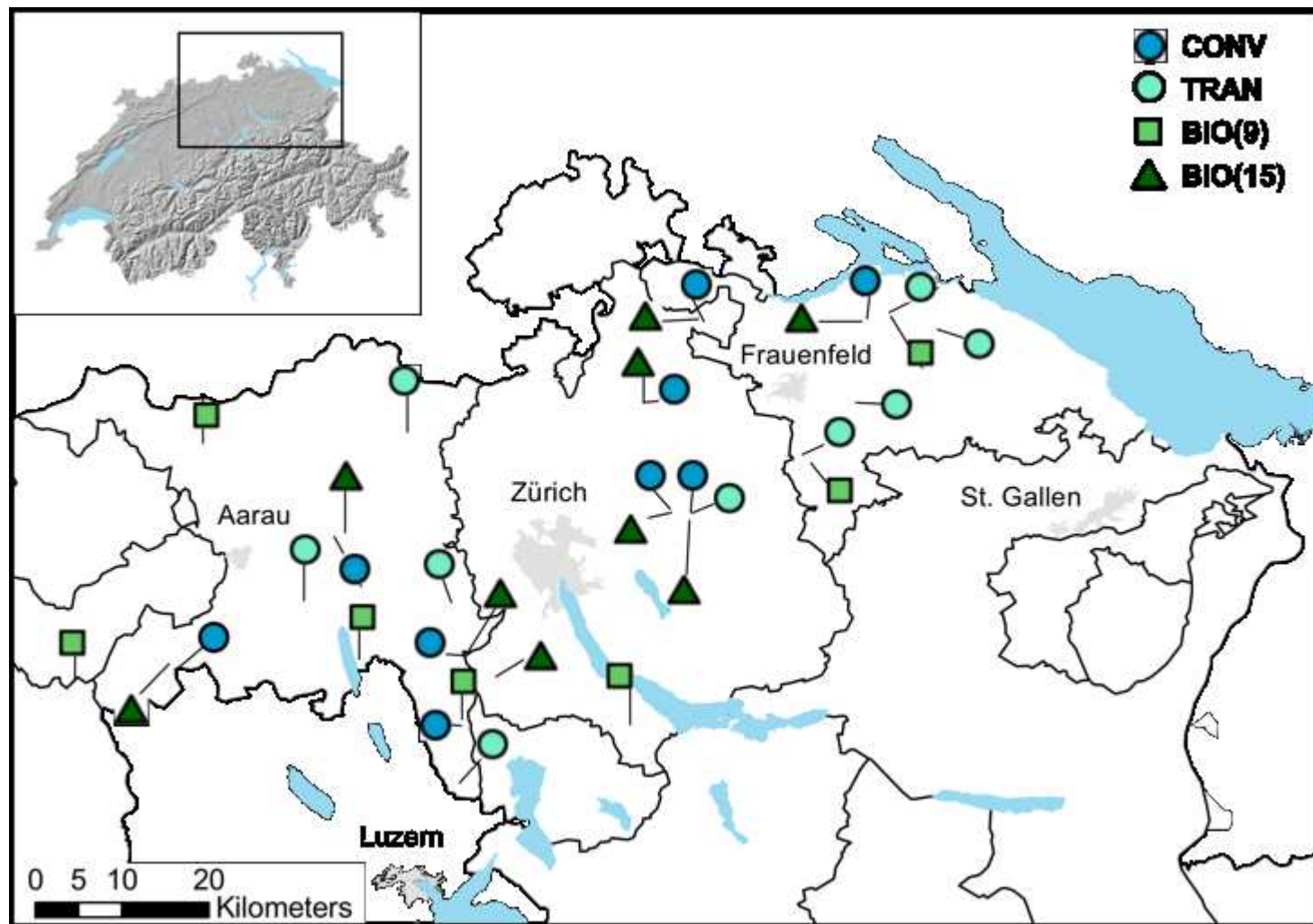
Fertilizer-N (kg N <sub>soluble</sub> /ha)	no	117.667 <b>a</b>	9.333 6	86.500 <b>ab</b>	16.506 6	61.600 <b>b</b>	5.418 5	60.400 <b>b</b>	12.995 5	8.792	<b>0.032</b>
Fertilizer-P (kg P <sub>2</sub> O <sub>5</sub> /ha)	no	26.833	14.554 6	75.333	14.033 6	61.200	9.404 5	66.800	7.493 5	6.618	0.085
Fertilizer-K (kg K <sub>2</sub> O/ha)	no	57.667 <b>a</b>	27.559 6	157.333 <b>ab</b>	22.317 6	187.200 <b>b</b>	21.386 5	210.200 <b>b</b>	24.132 5	11.396	<b>0.010</b>

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**Table 3.** For the multifunctionality assessment, individual parameters (indicator variables) were assigned to five ecosystem function (EF) categories, namely: “productivity”, “soil fertility”, “soil microbial activity”, “biodiversity conservation”, and “potential biocontrol of pests”. For each indicator variable it is indicated whether it was assessed for one or both crops (M= maize, W=wheat). \*Weed cover was defined as a disfunction because it is an undesirable aspect for farmers.

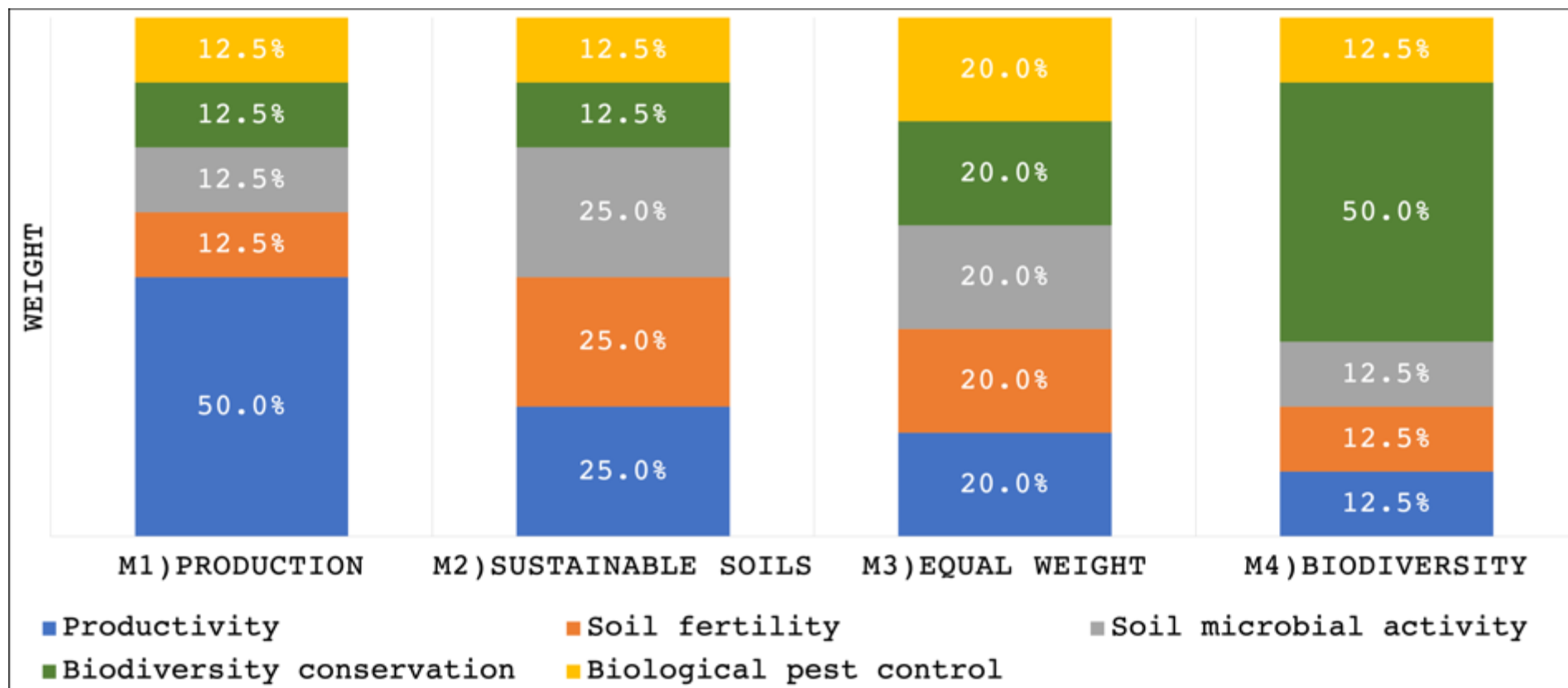
Ecosystem function (EF) category	Indicator variable	Crop	
Productivity	Yield	M, W	956
	Weed cover *	M, W	
Soil fertility	C <sub>org</sub>	W	957
	Soil P	W	958
	Soil K	W	
	Soil Mg	W	
Soil microbial activity	AMF colonization	M, W	959
	Microbial biomass C	W	
	Soil Respiration	W	
Biodiversity conservation	Weed species richness ( <i>S</i> )	M, W	960
	Weed species evenness ( <i>J</i> )	M, W	
	Spider species richness ( <i>S</i> )	M	
	Spider species evenness ( <i>J</i> )	M	961
	OTU richness of root associated fungi ( <i>S</i> )	W	
	OTU evenness of root associated fungi ( <i>J</i> )	W	
Potential biocontrol of pests	Spider abundance	M	

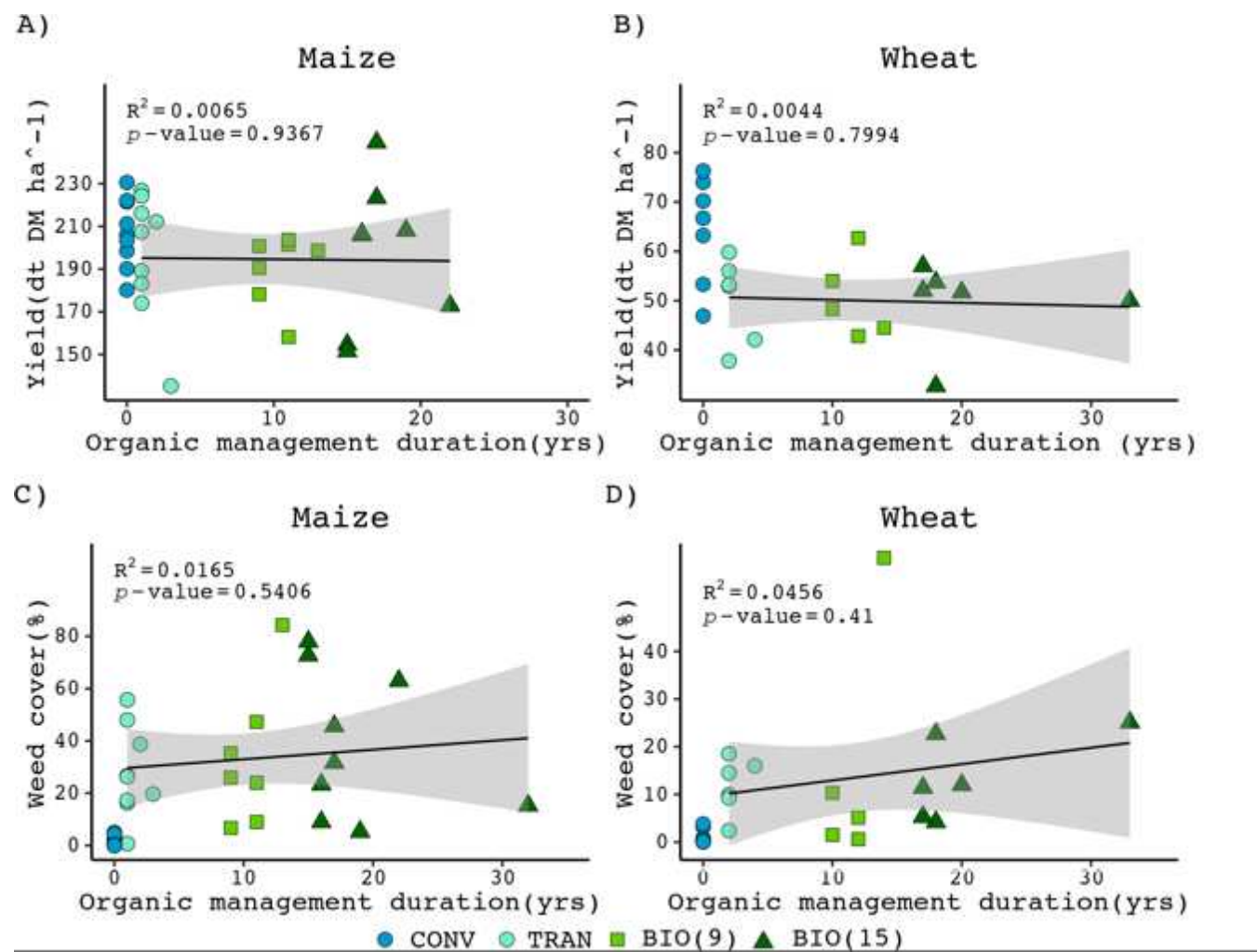
962 Fig. 1.

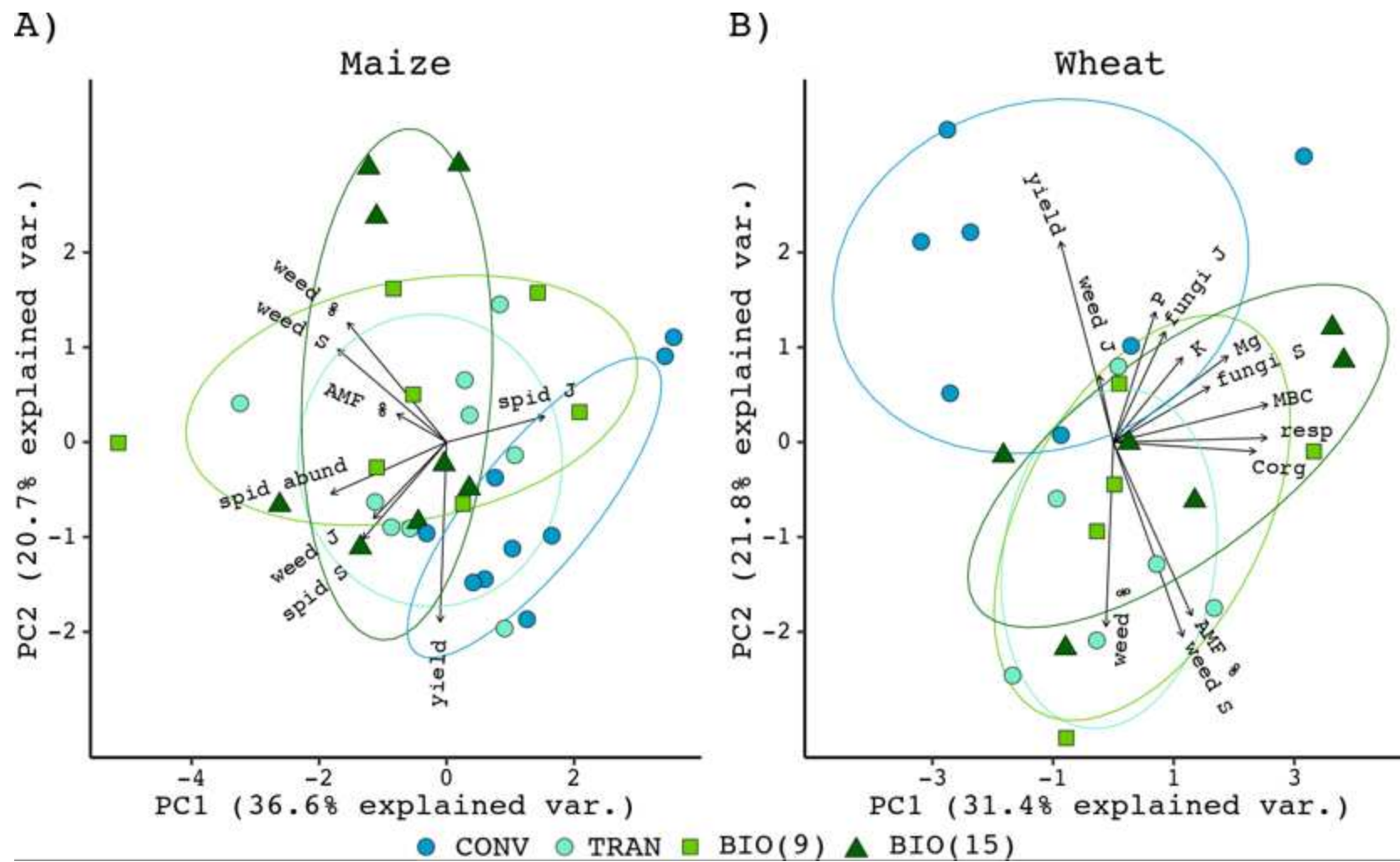


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964 **Fig. 2.**







971 **Fig.5.**

